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For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: **METHOD AND NUCLEIC ACIDS FOR THE ANALYSIS OF BREAST CELL PROLIFERATIVE DISORDERS**

(57) Abstract: The present invention relates to modified and genomic sequences, to oligonucleotides and/or PNA-oligomers for detecting the cytosine methylation state of genomic DNA, as well as to a method for ascertaining genetic and/or epigenetic parameters of genes for use in the differentiation, diagnosis, treatment and/or monitoring of breast cell proliferative disorders, or the predisposition to breast cell proliferative disorders.

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INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 03/07827

A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 C12Q1/68

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C12Q

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, PAJ, EMBASE, Sequence Search, BIOSIS, MEDLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	WO 02/059347 A (UNIV JOHNS HOPKINS MED) 1 August 2002 (2002-08-01) claims 1-44	1,2, 12-38
Y	WO 02/38801 A (GRIGG GEOFFREY WALTER ;MOLLOY PETER (AU); HUMAN GENETIC SIGNATURES) 16 May 2002 (2002-05-16) claims 1-16 ----- -/--	1,2, 12-38

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- "Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- "&" document member of the same patent family

Date of the actual completion of the international search

11 February 2004

Date of mailing of the international search report

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INTERNATIONAL SEARCH REPORT

International Application No
PCT/EP 03/07827

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	<p>GITAN RAAD S ET AL: "Methylation-specific oligonucleotide microarray: a new potential for high-throughput methylation analysis"</p> <p>GENOME RESEARCH, COLD SPRING HARBOR LABORATORY PRESS, US, vol. 12, no. 1, January 2002 (2002-01), pages 158-164, XP002208051 ISSN: 1088-9051 the whole document</p>	1,2, 12-38
Y	<p>-----</p> <p>YAN P S ET AL: "Dissecting complex epigenetic alterations in breast cancer using CpG Island microarrays"</p> <p>CANCER RESEARCH, AMERICAN ASSOCIATION FOR CANCER RESEARCH, BALTIMORE, MD, US, vol. 61, no. 23, 1 December 2001 (2001-12-01), pages 8375-8380, XP002243660 ISSN: 0008-5472 the whole document</p>	1,2, 12-38
Y	<p>-----</p> <p>HUANG TIM HUI-MING ET AL: "Methylation profiling of CpG islands in human breast cancer cells"</p> <p>HUMAN MOLECULAR GENETICS, OXFORD UNIVERSITY PRESS, SURREY, GB, vol. 8, no. 3, March 1999 (1999-03), pages 459-470, XP002175857 ISSN: 0964-6906 the whole document</p>	1,2, 12-38
Y	<p>-----</p> <p>DE IORIO P ET AL: "Implication of laminin and collagen type IV expression in the progression of breast carcinoma."</p> <p>ANTICANCER RESEARCH. GREECE 2001 MAR-APR, vol. 21, no. 2B, March 2001 (2001-03), pages 1395-1399, XP008027518 ISSN: 0250-7005 the whole document</p>	1,2, 12-38
X	<p>-----</p> <p>WO 01/40521 A (CURAGEN CORP ; LEACH MARTIN (US); SHIMKETS RICHARD A (US)) 7 June 2001 (2001-06-07) Human coding sequence polymorphic site SEQ ID NO: 544. Claim 1; Page 260; SEQ ID NO: 542. Claim 1; Page 260; SEQ ID NO: 540. Claim 1; Page 259 page 260; claim 1</p>	4-11
X	<p>-----</p> <p>WO 01/29262 A (POHL MARK ; ORCHID BIOSCIENCES INC (US); PICOULT NEWBURG LESLIE (US)) 26 April 2001 (2001-04-26) SNP specific lower PCR primer SEQ ID 3530. Claim 1; Page 68claim 1</p> <p style="text-align: center;">----- -/--</p>	3-11

INTERNATIONAL SEARCH REPORT

International Application No
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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 01/77373 A (PIEPENBROCK CHRISTIAN ;BERLIN KURT (DE); EPIGENOMICS AG (DE); OLEK) 18 October 2001 (2001-10-18) the whole document -----	3-11
P,X	WO 02/070742 A (EPIGENOMICS AG) 12 September 2002 (2002-09-12) the whole document -----	1-38
Y	US 5 744 305 A (FODOR STEPHEN P A ET AL) 28 April 1998 (1998-04-28) cited in the application the whole document -----	12

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 03/07827

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.: 39
because they relate to subject matter not required to be searched by this Authority, namely:
see FURTHER INFORMATION sheet PCT/ISA/210
2. ☐ Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☒ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Invention 1: claims 1-38 (all partially)

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.1

Claims 24 - 35 include the step of obtaining a biological sample containing genomic DNA and are therefore directed to a diagnostic method practised on the human/animal body. The search has been carried out and based on the differential methylation state of genes in breast cell proliferative disorders and the detection thereof.

Continuation of Box I.1

Claims Nos.: 39

Rule 39.1(iv) PCT - Method for treatment of the human or animal body by therapy

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

Invention 1: claims 1-38 (all partially)

Isolated nucleic acids, nucleic acids or peptide nucleic acids oligomers derived from SEQ ID NO:1 (COL4A2 gene), sets thereof and arrays or kits comprising them for the use in methods for differentiating between breast cell proliferative disorders and normal tissue based on the methylation state.

Invention 2-73: claims 1-38 (all partially)

Idem as invention 1 but limited to the sequences having SEQ ID NO:2-73 (Invention 2 is limited to SEQ ID NO:2; Invention 3 is limited to SEQ ID NO:3; ...; Invention 73 is limited to SEQ ID NO:73)

Invention 74: claims 1-38 (all partially)

Isolated nucleic acids, nucleic acids or peptide nucleic acids oligomers derived from SEQ ID NO:366 (Calcitonin gene), sets thereof and arrays or kits comprising them for the use in methods for differentiating between breast cell proliferative disorders and normal tissue based on the methylation state.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 03/07827

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